Blood Viscosity in Subjects With Normoglycemia and Prediabetes

Concetta Irace, Claudio Carallo, Faustina Scavelli, Maria Serena De Franceschi, Teresa Esposito, and Agostino Gnasso

OBJECTIVE

Blood viscosity (BV) is higher in diabetic patients and might represent a risk factor for the development of insulin resistance and type 2 diabetes. However, data in subjects with normal glucose or prediabetes are missing. In the current study, we evaluated the relationship between BV and blood glucose in subjects with normal glucose or prediabetes.

RESEARCH DESIGN AND METHODS

Enrolled subjects were divided into three groups according to blood glucose: group A (*n* = 74), blood glucose <90 mg/dL; group B (*n* = 96), blood glucose ranging from 90 to 99 mg/dL; and group C (*n* = 94), blood glucose ranging from 100 to 125 mg/dL. BV was measured at 37°C with a cone-plate viscometer at shear rates ranging from 225 to 22.5 s^{-1}.

RESULTS

Blood pressure, blood lipids, fibrinogen, and plasma viscosity were similar in the three groups. BMI and waist circumference were significantly increased in group C. Hematocrit ($P < 0.05$) and BV (P between 0.01 and 0.001) were significantly higher in groups B and C compared with group A. Blood glucose was significantly and inversely correlated with HDL cholesterol and directly with BMI, waist, hematocrit ($r = 0.134$), and BV (from 225 s^{-1} to 22.5 s^{-1}; r ranging from 0.162 to 0.131). BV at shear rate 225 s^{-1} was independently associated with blood glucose.

CONCLUSIONS

The current study shows a direct relationship between BV and blood glucose in nondiabetic subjects. It also suggests that, even within glucose values considered completely normal, individuals with higher blood glucose levels have increased BV comparable with that observed in subjects with prediabetes.

Dipartimento di Medicina Sperimentale e Clinica "G. Salvatore," "Magna Græcia" University, Catanzaro, Italy

Corresponding author: Agostino Gnasso, gnasso@unicz.it.

Blood viscosity (BV) is the force that counteracts the free sliding of the blood layers within the circulation and depends on the internal cohesion between the molecules and the cells. Abnormally high BV can have several negative effects: the heart is overloaded to pump blood in the vascular bed, and the blood itself, more viscous, can damage the vessel wall. Furthermore, according to Poiseuille's law (1), BV is

inversely related to flow and might therefore reduce the delivery of insulin and glucose to peripheral tissues, leading to insulin resistance or diabetes (2–5).

It is generally accepted that BV is increased in diabetic patients (6–8). Although the reasons for this alteration are still under investigation, it is believed that the increase in osmolarity causes increased capillary permeability and, consequently, increased hematocrit and viscosity (9). It has also been suggested that the osmotic diuresis, consequence of hyperglycemia, could contribute to reduce plasma volume and increase hematocrit (10).

Cross-sectional studies have also supported a link between BV, hematocrit, and insulin resistance (11–17). Recently, a large prospective study has demonstrated that BV and hematocrit are risk factors for type 2 diabetes. Subjects in the highest quartile of BV were >60% more likely to develop diabetes than their counterparts in the lowest quartile (18). This finding confirms previous observations obtained in smaller or selected populations, in which the association between hemoglobin or hematocrit and occurrence of type 2 diabetes was investigated (19–22).

These observations suggest that the elevation in BV may be very early, well before the onset of diabetes, but definite data in subjects with normal glucose or prediabetes are missing. In the current study, we evaluated the relationship between BV and blood glucose in subjects with normal glucose or prediabetes in order to verify whether alterations in viscosity are appreciable in these subjects and at which blood glucose concentration they appear.

RESEARCH DESIGN AND METHODS

Subjects were enrolled among free-living participants in a cardiovascular disease prevention program between February 2011 and December 2012 (23,24). In order to reduce the influence of confounding factors, cigarette smoking, diabetes, plasma triglycerides >400 mg/dL, female sex before menopause, and drug use (chronic treatment and any drug in the week before blood withdrawal) were exclusion criteria for the present analysis.

According to blood glucose levels, participants were divided into three groups: group A, blood glucose <90 mg/dL; group B, blood glucose between 90 and 99 mg/dL; and group C, blood glucose between 100 and 125 mg/dL.

The ethics committee of Azienda Policlinico Mater Domini approved the study. All recruited subjects gave informed consent.

All subjects included in the study underwent a complete clinical examination and blood withdrawal. Standing height without shoes was measured to the nearest 0.5 cm. Weight was measured to the nearest 0.1 kg in ordinary street clothes. BMI was computed as weight (in kg) divided by height (in m^2). Waist circumference was measured midway between the lower rib margin and the iliac crest. Systolic and diastolic blood pressure was measured, on the right arm, after the participant had been resting for at least 5 min, with a standardized sphygmomanometer. Cigarette smoking and ongoing drug therapies were investigated by questionnaire.

Venous blood for routine and viscosity analyses was collected in the morning before the breakfast after overnight fasting in order to avoid the influence of postprandial lipid increase on hemorheological parameters (25).

Attention was paid to avoid venous stasis, and the hemostatic loop, when used, was immediately removed after cannulation of the vein. Blood glucose and lipids were measured by routine methods. Subjects with blood glucose <100 mg/dL were classified as nondiabetic, those with values between 100 and 125 mg/dL were classified as prediabetic, and those with blood glucose >125 mg/dL were classified as diabetic and excluded from the present analysis.

Blood and plasma viscosity were measured within 2 h from blood withdrawal; the blood specimen was added with heparin (35 IU/mL). Viscosity measurement was performed at 37°C with a cone-plate viscometer (Wells-Brookfield DV-III; Wells-Brookfield, Stoughton, MA) equipped with a cp-40 spindle. BV was recorded at shear rates ranging from 225 to

22.5 s^{-1}. For plasma viscosity, the average of measurements at shear rates of 225 and 90 s^{-1} was calculated. The coefficient of variation for blood and plasma viscosity was <3%. Microhematocrit was measured without correction for plasma trapping. The coefficient of variation for microhematocrit was ~1%.

Erythrocyte rigidity (Tk) was evaluated according to the Dintenfass formula: Tk = $(\mu r^{0.4} - 1) \cdot (\mu r^{0.4} \cdot h)^{-1}$, where h is hematocrit and μr is the relative BV (BV/plasma viscosity).

Statistical Analysis

All statistical analyses were performed by SPSS 17.0 for Windows. All studied variables except triglycerides had normal distribution. Triglycerides were log transformed before analyses. One-way ANOVA and LSD post hoc test were used to test the difference in studied variables between groups with different blood glucose concentrations. Pearson correlation coefficient was used to test the correlation between continuous variables. Multiple linear regression analysis was performed to evaluate the independent association of clinical and biochemical variables with blood glucose.

RESULTS

The clinical, biochemical, and hemorheological characteristics of the 264 participants are shown in Table 1. Seventy-four subjects had blood glucose <90 mg/dL (group A), 96 subjects had blood glucose ranging from 90 to 99 mg/dL (group B), and 94 had blood glucose ranging from 100 to 125 mg/dL (group C). Blood pressure, blood lipids, fibrinogen, and plasma viscosity were similar in the three groups. BMI and waist were significantly increased in subjects with prediabetes (group C) compared with the other two groups ($P < 0.05$). Subjects with high normal blood glucose were younger ($P < 0.05$) compared with the other two groups. Hematocrit ($P < 0.05$) and BV (P between 0.01 and 0.001) were significantly higher in subjects with prediabetes and in those with blood glucose ranging from 90 to 99 mg/dL compared with subjects with blood glucose <90 mg/dL. Tk was similar in the three groups.

Table 1—Clinical, biochemical, and rheological characteristics of subjects

	Group A	Group B	Group C	*P*
n	74	96	94	
Age (years)	56.2 ± 12.4	52.8 ± 10.8	57.2 ± 10.0	≤0.02
Cholesterol (mg/dL)	209 ± 41	220 ± 40	213 ± 40	≤0.23
Triglycerides (mg/dL)	116 ± 51	132 ± 64	141 ± 74	≤0.09
HDL cholesterol (mg/dL)	53 ± 17	51 ± 15	49 ± 12	≤0.27
Glucose (mg/dL)	85 ± 5	95 ± 3	108 ± 7	—
BMI (kg/m^2)	28.3 ± 4.1	28.0 ± 4.5	29.9 ± 4.9	≤0.01
Waist (cm)	91 ± 10	91 ± 13	96 ± 10	≤0.01
SBP (mmHg)	139 ± 22	136 ± 19	139 ± 24	≤0.60
DBP (mmHg)	86 ± 10	85 ± 10	87 ± 10	≤0.59
Hematocrit (%)	42.6 ± 3.9	44.0 ± 3.5	43.8 ± 3.4	≤0.05
BV_{225} (cP)	4.58 ± 0.60	4.98 ± 0.73	4.90 ± 0.72	≤0.01
BV_{90} (cP)	5.38 ± 0.76	5.95 ± 1.07	5.79 ± 0.99	≤0.01
BV_{45} (cP)	6.08 ± 1.01	6.94 ± 1.82	6.78 ± 1.58	≤0.01
$BV_{22.5}$ (cP)	7.40 ± 1.35	8.61 ± 2.87	8.41 ± 2.51	≤0.01
Plasma viscosity (cP)	1.48 ± 0.15	1.54 ± 0.16	1.55 ± 0.16	≤0.06
Fibrinogen (mg/dL)	323 ± 86	337 ± 85	318 ± 82	≤0.33
Tk	0.85 ± 0.06	0.85 ± 0.08	0.83 ± 0.07	≤0.18

Group A: blood glucose <90 mg/dL; group B: blood glucose 90–99 mg/dL; group C: blood glucose 100–125 mg/dL. DBP, diastolic blood pressure; SBP, systolic blood pressure.

In simple correlation analysis in the whole population, blood glucose was significantly and inversely correlated with HDL cholesterol ($r = -0.123$) and directly with BMI ($r = 0.142$), waist ($r = 0.180$), hematocrit ($r = 0.134$), and BV (from shear rate 225 s^{-1} to shear rate 22.5 s^{-1}; r ranging from 0.162 to 0.131). No correlation was found with age, total cholesterol, triglycerides, plasma viscosity, fibrinogen, or Tk. All the above variables significantly correlated with blood glucose, as well as age significantly different between groups at ANOVA, were tested for independent association with blood glucose in stepwise multiple regression analyses. (Age, HDL cholesterol, BMI, waist, and hematocrit were constantly in the model, while the viscosities at different shear rates were introduced one at a time.) Only BV at shear rate 225 s^{-1} resulted independently associated with blood glucose, while BV at lower shear rates was not (Table 2).

Two additional stepwise multiple regression analyses were performed to identify the variables independently associated with the viscosity at a high shear rate (225 s^{-1}) and low shear rate (22.5 s^{-1}). All variables significantly correlated to these two viscosities were included as independent variables (for BV at shear rate 225 s^{-1} [correlation coefficients], age [−0.178], total cholesterol [0.258], triglycerides [0–185], HDL cholesterol [−0.202], blood glucose [0.162], hematocrit [0.440], plasma viscosity [0.438], and Tk [0.294]; for BV at shear rate 22.5 s^{-1}, age [−0.148], total cholesterol [0.219], hematocrit [0.207], plasma viscosity

[0.355], and Tk [0.377]). The results indicate that hematocrit, plasma viscosity, and erythrocyte deformability are the only variables independently associated with the viscosity in these subjects.

CONCLUSIONS

The current study shows, for the first time, a direct relationship between BV and blood glucose in nondiabetic subjects. It also suggests that, even within glucose values considered completely normal, individuals with higher blood glucose levels have increases in BV comparable with those observed in subjects with prediabetes.

Many studies thus far have investigated BV in patients with diabetes (26). Usually, however, these studies were performed in small groups of patients, often only at few shear rates, and sometimes did not demonstrate any difference between diabetic and control subjects (27,28). Some findings have suggested a role for the increase in BV as a pathogenetic factor for the development of microvascular complications (29,30). Important increases in BV have been reported in diabetic retinopathy, and it has been hypothesized that these changes lead to a prolonged reduction in the supply of oxygen and nutrients to the capillaries, causing the development of angiopathy (31,32). Once retinopathy has developed, its progression may be favored by a reduction in BV and hemoglobin (33). Overall, changes in viscosity in diabetic patients are accepted as common and as a result of the disease. However, the relationship between blood glucose, diabetes, and viscosity may be much more complex. First, it is useful to underline that the blood is a non-Newtonian fluid, in that its viscosity varies with the flow velocity. Specifically, when the flow velocity is high, the cellular component of blood (red cells, white cells, and platelets) is concentrated at the center of the vessel, while the plasma flows in the periphery. In conditions of low flow velocities, the cellular component occupies the entire column of flowing blood, causing an increase in viscosity. BV at high shear rates (≥90 s^{-1}, corresponding with in vivo systolic flow condition) is strongly

Table 2—Stepwise multiple regression analysis

	β-Coefficient	*t*	*P*
BV 225 s^{-1}	2.421	2.179	0.031
Variables not in the model			
Age	0.144	1.870	0.063
HDL cholesterol	−0.109	−1.386	0.168
BMI	0.055	0.718	0.474
Waist	0.096	1.256	0.211
Hematocrit	0.061	0.711	0.478

Dependent variable: blood glucose.

influenced by erythrocyte deformability, while at lower shear rates (representing in vivo diastolic flow condition) red cell aggregation plays the most important role (34). The deformability and the aggregation of erythrocytes are, in turn, driven by different characteristics of the erythrocytes and the plasma. Furthermore, factors such as cigarette smoking, hyperlipidemia, and hyperfibrinogenemia, which are very frequent in patients with diabetes, may alter BV. In light of this, it is evident that further studies are needed to clarify the relationship between diabetes and BV.

The importance of BV was recently highlighted after the observation that it increases the risk of developing diabetes in normoglycemic subjects participating in the Atherosclerosis Risk in Communities (ARIC) study (18). In that study, the authors did not directly measure BV but used two validated formulas: one for the shear rate 208 s^{-1} and the other for the shear rate 0.5 s^{-1} (35). The findings of our study are in line with the conclusions of the ARIC Study and further demonstrate that the BV of the subjects with prediabetes or high normal blood glucose is significantly higher compared with that of subjects with low normal blood glucose at all considered shear rates. This supports the hypothesis that the alterations are very early and present in any flow condition. Furthermore, these findings are also in line with the results of a previous study, which showed that the BV in patients with prediabetes was comparable with that of diabetic subjects with complications (36) and significantly higher than that of healthy control subjects. If increased viscosity somehow alters the supply of oxygen and nutrients to the tissues, then the observed changes may contribute to the development of peripheral insulin resistance (by reduced glucose utilization in the muscle) and, in the long term, diabetes. It cannot be excluded that a slight increase in blood glucose, secondary to other conditions of insulin resistance such as aging, inflammation, physical inactivity, etc., can cause increased BV, thereby creating a vicious circle. Clearly, this requires further study for a more exact definition.

The observation that the change in viscosity is already evident at blood glucose values >90 mg/dL seems very important. This allows understanding of why in some studies, no difference was observed between patients with diabetes and control subjects (27,28). Probably, the control subjects had high normal blood glucose levels or were prediabetic. In a previous study, conducted in a small population of diabetic patients and a control group, we found no difference in BV measured at shear rate of 225 s^{-1} (37). The control subjects, however, had a mean blood glucose of >95 mg/dL, and although nondiabetic, they probably already had increased BV. Furthermore, the value of 90 mg/dL would suggest a greater attention in subjects who exceed this threshold in terms of correction of the conditions that can cause increased viscosity.

In this regard, it is intriguing to try to identify the factors that influence the viscosity. Changes in osmolality or osmotic diuresis have been suggested as factors able to modify BV (9,10). While these factors may play a role at very high blood glucose concentrations, it seems unlikely that they may affect BV within normal or slightly increased blood glucose values. Among the variables considered in our study, hematocrit, plasma viscosity, and erythrocyte deformability are the factors independently associated with high and low shear rates BV, confirming recently published data (38). The gradual increase in plasma viscosity, hematocrit, and Tk, even within levels considered completely normal, leads to increased BV. It is possible to hypothesize that gradually increasing concentrations of blood glucose can directly affect the erythrocytes, possibly resulting in impaired deformability and aggregation. If confirmed in larger and prospective studies, these results could indicate the need for very early intervention on these factors in an attempt to prevent the development of diabetes.

The plasma viscosity was slightly higher in groups B and C compared with group A but only close to statistical significance. The plasma viscosity is strongly influenced by the levels of

fibrinogen and other plasma proteins and, to a lesser extent, by obesity and blood lipids (38,39). In the current study, subjects with high normal blood glucose or prediabetes have higher values of BMI and slightly of blood lipids, and this may explain why plasma viscosity tends to higher values, thus contributing to the increase in whole BV.

The current study has, in our opinion, some limitations. Insulin resistance has not been evaluated, since insulin was not measured. Similarly, data on inflammation and plasma proteins are lacking, although the careful selection of the subjects, with the exclusion of those taking drugs, may have limited the impact of these factors. The subjects were classified on the basis of a single fasting glucose, and HbA_{1c} was not available in this population; therefore, it is possible that a few subjects were classified inappropriately. We believe that this potential bias might have affected only a few subjects and therefore does not invalidate the results. Finally, the erythrocyte deformability was calculated and not directly measured. Therefore, the data on erythrocyte deformability should be interpreted with caution.

In conclusion, the main finding of the study is that BV significantly increases already at high-normal blood glucose levels, independently of other common determinants of hemorheology. Intervention studies are needed to verify whether changes in BV can influence the development of type 2 diabetes.

References

1. Guyton A, Hall J. *Textbook of Medical Physiology*. 10th ed. Philadelphia, PA, WB Saunders Company, 2000, p. 212–213

2. Stuart J, Kenny MW. Blood rheology. J Clin Pathol 1980;33:417–429

3. Yang YJ, Hope ID, Ader M, Bergman RN. Insulin transport across capillaries is rate limiting for insulin action in dogs. J Clin Invest 1989;84:1620–1628

4. Schultz TA, Lewis SB, Westbie DK, Wallin JD, Gerich JE. Glucose delivery: a modulator of glucose uptake in contracting skeletal muscle. Am J Physiol 1977;233:E514–E518

5. Baron AD, Clark MG. Role of blood flow in the regulation of muscle glucose uptake. Annu Rev Nutr 1997;17:487–499

6. Skovborg F, Nielsen AV, Schlichtkrull J, Ditzel J. Blood-viscosity in diabetic patients. Lancet 1966;1:129–131

7. Dintenfass L. Blood viscosity factors in severe nondiabetic and diabetic retinopathy. Biorheology 1977;14:151–157

8. Schmid-Schönbein H, Volger E. Red-cell aggregation and red-cell deformability in diabetes. Diabetes 1976;25(Suppl. 2):897–902

9. Meiselman HJ, Merrill EW, Gilliland ER, Pelletier GA, Salzman EW. Influence of plasma osmolarity on the rheology of human blood. J Appl Physiol 1967;22:772–781

10. Lowe GD, Lowe JM, Drummond MM, et al. Blood viscosity in young male diabetics with and without retinopathy. Diabetologia 1980;18:359–363

11. Høieggen A, Fossum E, Moan A, Enger E, Kjeldsen SE. Whole-blood viscosity and the insulin-resistance syndrome. J Hypertens 1998;16:203–210

12. Nordby G, Moan A, Kjeldsen SE, Os I. Relationship between hemorheological factors and insulin sensitivity in normotensive and hypertensive premenopausal women. Am J Hypertens 1995;8:439–444

13. Catalano C, Muscelli E, Natali A, et al. Reciprocal association between insulin sensitivity and the haematocrit in man. Eur J Clin Invest 1997;27:634–637

14. Smith S, Julius S, Jamerson K, Amerena J, Schork N. Hematocrit levels and physiologic factors in relationship to cardiovascular risk in Tecumseh, Michigan. J Hypertens 1994;12:455–462

15. Facchini FS, Carantoni M, Jeppesen J, Reaven GM. Hematocrit and hemoglobin are independently related to insulin resistance and compensatory hyperinsulinemia in healthy, non-obese men and women. Metabolism 1998;47:831–835

16. Capoğlu I, Unüvar N, Bektaş Y, Yilmaz O, Kaya MD. The effects of high haematocrit levels on glucose metabolism disorders. J Int Med Res 2002;30:433–437

17. Pérez-Martin A, Dumortier M, Pierrisnard E, Raynaud E, Mercier J, Brun JF. Multivariate analysis of relationships between insulin sensitivity and blood rheology: is plasma viscosity a marker of insulin resistance? Clin Hemorheol Microcirc 2001;25:91–103

18. Tamariz LJ, Young JH, Pankow JS, et al. Blood viscosity and hematocrit as risk factors for type 2 diabetes mellitus: the atherosclerosis risk in communities (ARIC) study. Am J Epidemiol 2008;168:1153–1160

19. Wannamethee SG, Perry IJ, Shaper AG. Hematocrit and risk of NIDDM. Diabetes 1996;45:576–579

20. Tulloch-Reid MK, Hanson RL, Saremi A, et al. Hematocrit and the incidence of type 2 diabetes in the pima indians. Diabetes Care 2004;27:2245–2246

21. Medalie JH, Papier CM, Goldbourt U, Herman JB. Major factors in the development of diabetes mellitus in 10,000 men. Arch Intern Med 1975;135:811–817

22. Wilson PW, McGee DL, Kannel WB. Obesity, very low density lipoproteins, and glucose intolerance over fourteen years: The Framingham Study. Am J Epidemiol 1981;114:697–704

23. Gnasso A, Calindro MC, Carallo C, et al. Awareness, treatment and control of hyperlipidaemia, hypertension and diabetes mellitus in a selected population of southern Italy. Eur J Epidemiol 1997;13:421–428

24. Irace C, Cortese C, Fiaschi E, et al. Components of the metabolic syndrome and carotid atherosclerosis: role of elevated blood pressure. Hypertension 2005;45:597–601

25. Stamos TD, Rosenson RS. Low high density lipoprotein levels are associated with an elevated blood viscosity. Atherosclerosis 1999;146:161–165

26. Cho YI, Mooney MP, Cho DJ. Hemorheological disorders in diabetes mellitus. J Diabetes Sci Tech 2008;2:1130–1138

27. Schnyder L, Walter R, Rohrer A, Contesse J, Reinhart WH. No influence of C-peptide, insulin, and glucagon on blood viscosity in vitro in healthy humans and patients with diabetes mellitus. Clin Hemorheol Microcirc 2001;24:65–74

28. Rimmer T, Fleming J, Kohner EM. Hypoxic viscosity and diabetic retinopathy. Br J Ophthalmol 1990;74:400–404

29. Merimee TJ. Diabetic retinopathy. A synthesis of perspectives. N Engl J Med 1990;322:978–983

30. Williamson TH, Rumley A, Lowe GD. Blood viscosity, coagulation, and activated protein C resistance in central retinal vein occlusion: a population controlled study. Br J Ophthalmol 1996;80:203–208

31. Chmiel B, Cierpka L. Organ preservation solutions impair deformability of erythrocytes in vitro. Transplant Proc 2003;35:2163–2164

32. Linderkamp O, Pöschl J, Ruef P. Blood cell deformation in neonates who have sepsis. NeoReviews 2006;7:e517–e523

33. Irace C, Scarinci F, Scorcia V, et al. Association among low whole blood viscosity, haematocrit, haemoglobin and diabetic retinopathy in subjects with type 2 diabetes. Br J Ophthalmol 2011;95:94–98

34. Kim S, Popel AS, Intaglietta M, Johnson PC. Effect of erythrocyte aggregation at normal human levels on functional capillary density in rat spinotrapezius muscle. Am J Physiol Heart Circ Physiol 2006;290:H941–H947

35. de Simone G, Devereux RB, Chien S, Alderman MH, Atlas SA, Laragh JH. Relation of blood viscosity to demographic and physiologic variables and to cardiovascular risk factors in apparently normal adults. Circulation 1990;81:107–117

36. Richards RS, Nwose EU. Blood viscosity at different stages of diabetes pathogenesis. Br J Biomed Sci 2010;67:67–70

37. Irace C, Carallo C, Crescenzo A, et al. NIDDM is associated with lower wall shear stress of the common carotid artery. Diabetes 1999;48:193–197

38. Irace C, Carallo C, Scavelli F, et al. Influence of blood lipids on plasma and blood viscosity. Clin Hemorheol Microcirc. 7 February 2013 [Epub ahead of print]

39. Vayá A, Simó M, Santaolaria M, Carrasco P, Corella D. Plasma viscosity and related cardiovascular risk factors in a Spanish Mediterranean population. Thromb Res 2007;120:489–495